IN THE CLAIMS

Claims 100, 112, and 117 have been amended. Claims 100, 102-107, 109-112, 114-117, and 119-128 are pending in the present application. The following is the status of the claims of the above-captioned application, as amended.

1-99 (Cancelled)

100. (Currently Amended) An isolated nucleic acid sequence encoding a naturally-occurring polypeptide having phospholipase B activity, selected from the group consisting of:

(a) a nucleic acid sequence encoding a polypeptide having an amino acid sequence which has at least 90% identity with amino acids 20 to 464 of SEQ ID NO: 2;

(b) a nucleic acid sequence having at least 90% homology with nucleotides 568 to 2045 of SEQ ID NO: 1; and

(c) a nucleic acid sequence which hybridizes under medium-high stringency conditions with (i) nucleotides 568 to 2045 of SEQ ID NO: 1, (ii) the cDNA sequence contained in nucleotides 568 to 2045 of SEQ ID NO: 1, or (iii) a complementary strand of (i) or (ii).

101. (Cancelled)

102. (Previously Presented) The nucleic acid sequence of claim 100, which encodes a polypeptide having an amino acid sequence which has at least 90% identity with amino acids 20 to 464 of SEQ ID NO: 2.

103. (Previously Presented) The nucleic acid sequence of claim 102, which encodes a polypeptide having an amino acid sequence which has at least 95% identity with amino acids 20 to 464 of SEQ ID NO: 2.

104. (Previously Presented) The nucleic acid sequence of claim 103, which encodes a polypeptide having an amino acid sequence which has at least 97% identity with amino acids 20 to 464 of SEQ ID NO: 2.

105. (Previously Presented) The nucleic acid sequence of claim 100, which encodes a polypeptide comprising the amino acid sequence of SEQ ID NO: 2.

106. (Previously Presented) The nucleic acid sequence of claim 100, which encodes a polypeptide consisting of the amino acid sequence of SEQ ID NO: 2, or a fragment thereof which has phospholipase B activity.

107. (Previously Presented) The nucleic acid sequence of claim 106, which encodes a polypeptide

consisting of amino acids 20 to 464 of SEQ ID NO: 2.

108. (Cancelled)

109. (Previously Presented) The nucleic acid sequence of claim 100, which has at least 90% homology with nucleotides 568 to 2045 of SEQ ID NO: 1.

110. (Previously Presented) The nucleic acid sequence of claim 109, which has at least 95% homology with nucleotides 568 to 2045 of SEQ ID NO: 1.

111. (Previously Presented) The nucleic acid sequence of claim 110, which has at least 97% homology with nucleotides 568 to 2045 of SEQ ID NO: 1.

112. (Currently Amended) The nucleic acid sequence of claim 100, ~~which has~~ comprising the nucleic acid sequence of nucleotides 568 to 2045 of SEQ ID NO: 1.

113. (Cancelled)

114. (Previously Presented) The nucleic acid sequence of claim 100, which hybridizes under medium-high stringency conditions with (i) nucleotides 568 to 2045 of SEQ ID NO: 1, (ii) the cDNA sequence contained in nucleotides 568 to 2045 of SEQ ID NO: 1, or (iii) a complementary strand of (i) or (ii).

115. (Previously Presented) The nucleic acid sequence of claim 114, which hybridizes under high stringency conditions with (i) nucleotides 568 to 2045 of SEQ ID NO: 1, (ii) the cDNA sequence contained in nucleotides 568 to 2045 of SEQ ID NO: 1, or (iii) a complementary strand of (i) or (ii).

116. (Previously Presented) The nucleic acid sequence of claim 100, contained in *E. coli* pPH6 as deposited with NRRL under accession number B-30142.

117. (Currently Amended) An isolated nucleic acid sequence encoding a naturally-occurring polypeptide having phospholipase B activity, said nucleic acid sequence obtained by (a) identifying a clone containing a nucleic acid sequence which hybridizes under medium-high stringency conditions with (i) nucleotides 568 to 2045 of SEQ ID NO. 1, (ii) the cDNA sequence contained in nucleotides 568 to 2045 of SEQ ID NO. 1, or (iii) a complementary strand of (i) or (ii); and (b) isolating the nucleic acid sequence encoding a polypeptide having phospholipase B activity from the clone.

118. (Cancelled)

119. (Previously Presented) The nucleic acid sequence of claim 117 obtained by (a) identifying a clone containing a nucleic acid sequence which hybridizes under high stringency conditions with (i) nucleotides 568 to 2045 of SEQ ID NO. 1, (ii) the cDNA sequence contained in nucleotides 568 to 2045 of SEQ ID NO. 1, or (iii) a complementary strand of (i) or (ii); and (b) isolating the nucleic acid sequence encoding a polypeptide having phospholipase B activity from the clone.

120. (Previously Presented) A nucleic acid construct comprising the nucleic acid sequence of claim 100 operably linked to one or more control sequences which direct the production of the polypeptide in a suitable expression host.

121. (Previously Presented) A recombinant expression vector comprising the nucleic acid construct of claim 120.

122. (Previously Presented) A recombinant host cell comprising the nucleic acid construct of claim 120.

123. (Previously Presented) A method for producing a polypeptide having phospholipase B activity comprising (a) cultivating a strain comprising the nucleic acid sequence of claim 100 under conditions suitable for producing the polypeptide; and (b) recovering the polypeptide.

124. (Previously Presented) A method for producing a polypeptide having phospholipase B activity comprising (a) cultivating the recombinant host cell of claim 122 under conditions suitable for production of the polypeptide; and (b) recovering the polypeptide.

125. (Previously Presented) A nucleic acid construct comprising a gene encoding a protein operably linked to a nucleic acid sequence encoding a signal peptide consisting of nucleotides 510 to 567 of SEQ ID NO. 1, wherein the gene is foreign to the nucleic acid sequence.

126. (Previously Presented) A recombinant expression vector comprising the nucleic acid construct of claim 125.

127. (Previously Presented) A recombinant host cell comprising the nucleic acid construct of claim 125.

128. (Previously Presented) A method for producing a protein comprising (a) cultivating the recombinant host cell of claim 127 under conditions suitable for production of the protein; and (b) recovering the protein.

I. The Rejection of Claims 100, 102-104, 109-111, 114-115, and 120-124 under 35 U.S.C. § 112, First Paragraph

Claims 100, 102-104, 109-111, 114-115, and 120-124 remain rejected under 35 U.S.C. § 112, first paragraph, "as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention" for the reasons of record. This rejection is respectfully traversed for the reasons of record and for additional reasons discussed below.

The present invention relates to isolated nucleic acid sequences encoding a naturally-occurring polypeptide having phospholipase B activity, selected from the group consisting of:

(a) a nucleic acid sequence encoding a polypeptide having an amino acid sequence which has at least 95% identity with amino acids 20 to 464 of SEQ ID NO: 2;

(b) a nucleic acid sequence having at least 95% homology with nucleotides 568 to 2045 of SEQ ID NO: 1; and

(c) a nucleic acid sequence which hybridizes under at least high stringency conditions with (i) nucleotides 568 to 2045 of SEQ ID NO: 1, (ii) the cDNA sequence contained in nucleotides 568 to 2045 of SEQ ID NO: 1, or (iii) a complementary strand of (i) or (ii).

The Office states that Applicant's arguments filed 11/26/04 have been fully considered but are not persuasive for several reasons described below.

The Office states:

> [T]he structural features on which Applicant relies do not distinguish between nucleic acids that are claimed, i.e. those which meet the structural limitations of the claims and encode a phospholipase B, from those which are not claimed, i.e. those which meet the structural limitations of the claims but do not encode a phospholipase B. It must be kept in mind that the claims are not limited to nucleic acids isolated from nature or nucleic acids encoding phospholipase B enzymes found in nature. The claims also embrace man-made variants of SEQ ID NO: 1 or variants of nucleic acids that encode SEQ ID NO: 2 that encode polypeptides differing from SEQ ID NO: 2. Amino acids changes can be made in SEQ ID NO: 2 that will render the polypeptide inactive as a phospholipase B. Presumably, changes could also be made that would not eliminate phospholipase B activity. The specification does not teach characteristics of the proteins encoded by each class, active vs. inactive, that would distinguish them from one another, or allow one of skill in the art to envision those readable on the claims.

Preliminarily, to further prosecution, Applicant has amended the claims to recite "a naturally-occurring polypeptide having phospholipase B activity", but reserve the right to file continuing applications on the former subject matter.

Applicant respectfully points out that that the claims recite "a polypeptide having phospholipase B activity" and describe on page 3, lines 18-24, of the specification an assay for determing phospholipase B activity. Consequently, polypeptides having no phospholipase B activity are irrelevant to the claimed invention. The structural features on which Applicant relies do distinguish between nucleic acids that are claimed, *i.e.*, those which meet the structural limitations of

the claims and encode a phospholipase B, from those which are not claimed, *i.e.*, those which meet the structural limitations of the claims but do not encode a phospholipase B because the polypeptide encoded by the nucleic acids must have phospholipase B activity, as recited in the preamble of claim 1.

The Office also states:

> Bork (1994) at page 397 teaches that whether one can predict protein function based upon amino acid sequence homology to a protein of known function depends upon the data at hand. Bork cautions that even in simple cases where there is strong sequence similarity to a protein of known function, sequence variation can have diverse consequences. ... Bork (1998) teaches that molecular characteristics such as enzymatic activity, interaction partners, and pathway context can be predicted from sequence comparisons only qualitatively, and most of the functional features of proteins (with no known function) cannot be predicted from sequence data. Bork (1998) lists other pitfalls in predicting protein function from sequence comparisons (page 707, col. 2 through page 710). The Berka declaration then provides evidence that for many naturally-occurring metabolic enzymes, and the nucleic acids that encode them, which share sequence similarity of 90% or higher, the proteins have the same enzymatic activity.

Again, Applicant respectfully points out that the claimed polypeptides must have phospholipase B activity, assayed as described on page 3, lines 18-24, of the specification.

The Office Action also states:

> Wilson does not address whether the function of proteins can be predicted by sequence comparison to proteins of known function as asserted in Applicant's reply and in the declaration, but whether the function of putative protein folds (SCOP domains, page 242; page 245, col. 2) found within a new protein can be predicted by sequence comparison to proteins of known function having a that protein fold. Also, as indicated on page 242, col. 2, when Wilson speaks of two proteins having the "same precise function," he does not mean that the proteins actually perform the same enzymatic reaction on the same substrates to produce the same products. For example, as defined by Wilson, alcohol dehydrogenase and homoserine dehydrogenase have the "same precise function", but these enzymes do not act on the same substrates or produce the same products. By Wilson's definition, cholinesterase (EC 3.1.1.8) and phospholipase B, i.e. lysophospholipase, (EC 3.1.1.5) share the same precise function, i.e. they hydrolyze carboxylic esters. ... If one had predicted the function of SEQ ID NO: 2 from sequence data available at the time the application was filed, one might have predicted it was a phospholipase C ..."

Again, Applicant respectfully points out that the claimed polypeptides must have phospholipase B activity. The function defined by Applicant's specification is phospholipase B activity, not phospholipase C activity. One of ordinary skill in the art would readily recognize following Applicant's specification that predicting the activity associated with an enzyme from sequence data without more is insufficient and requires the additional step of actually assaying the enzyme for phospholipase B activity, as Applicant has done.

The Office Action also states:

> [T]he Office agrees (and has never disagreed) with the argument that if a naturally-occurring enzyme or naturally-occurring enzyme coding nucleotide sequence shares 90% sequence identity with SEQ ID NO: 2 or 1, respectively, or if a naturally-occurring enzyme coding nucleotide sequence hybridizes under medium-high stringency conditions to SEQ ID NO: 1, then more likely than not, the enzyme would

be a phospholipase B. However, the issue is whether the specification adequately describes such naturally occurring sequences and whether Applicant was in possession of such.

The specification fails to provide an adequate written description of the claimed genus of nucleic acid sequences for two reasons relevant to naturally occurring sequence. First, the claims are not limited to naturally-occurring nucleic acids or nucleic acids encoding a naturally-occurring phospholipase B. ... Second the only naturally-occurring nucleotide and amino acid sequences disclosed in the specification are SEQ ID NOs: 1 and 2. There is no evidence of record that Applicant was in possession of any other naturally-occurring sequences.

The Office asserts that the claims are not limited to naturally-occurring nucleic acids or nucleic acids encoding a naturally-occurring phospholipase B. Applicant has amended the claims to recite "a naturally-occurring polypeptide having phospholipase B activity".

The Office also asserts that the only naturally-occurring nucleotide and amino acid sequences disclosed in the specification are SEQ ID NOs: 1 and 2 and there is no evidence of record that Applicant was in possession of any other naturally-occurring sequences.

An essential feature of the claimed invention is that the isolated nucleic acid sequence hybridizes under medium-high or high stringency conditions to (i) nucleotides 568 to 2045 of SEQ ID NO: 1, (ii) the cDNA sequence contained in nucleotides 568 to 2045 of SEQ ID NO: 1, or (iii) a complementary strand of (i) or (ii), and encodes a polypeptide having phospholipase B activity. Applicant submits that a person of ordinary skill in the art would not expect substantial variation among species encompassed within the scope of the claims because the medium-high and high stringent hybridization conditions as set forth in the claims yield structurally similar DNAs. Thus a representative number of species is disclosed, since the hybridization conditions in combination with the coding function of DNA and the level of skill and knowledge in the art are adequate to show that Applicant was in possession of the claimed invention.

Another essential feature of the claimed invention is that the isolated nucleic acid sequence encodes a polypeptide having phospholipase B activity and has an amino acid sequence which has at least 90% identity with amino acids 20 to 464 of SEQ ID NO: 2. Applicant submits that a person of ordinary skill in the art would not expect substantial variation among species encompassed within the scope of the claims because the isolated nucleic acid sequence encoding a polypeptide having phospholipase B activity and having an amino acid sequence which has at least 90% identity with amino acids 20 to 464 of SEQ ID NO: 2 as set forth in the claims yield structurally similar DNAs. Thus a representative number of species is disclosed, since at least 90% identity in combination with the coding function of DNA and the level of skill and knowledge in the art are adequate to show that Applicant was in possession of the claimed invention.

A further essential feature of the claimed invention is that the isolated nucleic acid sequence encoding a polypeptide having phospholipase B activity has at least 90% homology with nucleotides 568 to 2045 of SEQ ID NO: 1. Applicant submits that a person of ordinary skill in the art would not expect substantial variation among species encompassed within the scope of the claims because the isolated nucleic acid sequence encoding a polypeptide having phospholipase B activity and having at

least 90% homology with nucleotides 568 to 2045 of SEQ ID NO: 1 as set forth in the claims yield structurally similar DNAs. Thus a representative number of species is disclosed, since the at least 90% homology in combination with the coding function of DNA and the level of skill and knowledge in the art are adequate to show that Applicant was in possession of the claimed invention.

Applicant submits, therefore, that the specification complies with the written description requirement and respectfully request reconsideration and withdrawal of the rejection.

II. The Rejection of Claims 100, 102-104, 109-111, 114-115, 117 and 119-124 under 35 U.S.C. § 112, First Paragraph

Claims 100, 102-104, 109-111, 114-115, 117 and 119-124 remain rejected under 35 U.S.C. § 112, first paragraph, because the specification, while being enabling for a nucleic acid sequence encoding phospholipase B wherein either the nucleic acid sequence comprises nucleotides 568 to 2045 of SEQ ID NO: 1 or the polypeptide comprises amino acids 20-464 of SEQ ID NO: 2, does not reasonably provide enablement for any other embodiments lying outside this scope for the reasons of record.

This rejection is respectfully traversed for the reasons of record and for additional reasons discussed below.

The reasoning provided by the Office for rejecting the claims is that the specification "does not demonstrate using a nucleic acid of SEQ ID NO: 1 to isolate a claimed nucleic acid from a different source, nor does the specification identify a source, from which one would be able to isolate a claimed nucleic acid, other than *A. oryzae*" and "the claims are not limited to sequences obtainable from a natural source, and the example does not teach how to make a nucleic acid readable on the claims that cannot be found in nature and encodes a different amino acid sequence than SEQ ID NO: 2."

Preliminarily, Applicant has amended the claims to recite "a naturally-occurring polypeptide having phospholipase B activity", as noted earlier.

The specification contains an extensive disclosure of techniques which are well known in the art and indeed routine for persons of ordinary skill in the art for making and using the claimed subject matter of the present invention. Applicant describes methods for preparing and probing DNA libraries (Example 1-2); for isolating nucleic acids encoding the phospholipases (Example 3); for determining cross-hybridization of the nucleic acids encoding phospholipases using (i) nucleotides 568 to 2045 of SEQ ID NO: 1, (ii) the cDNA sequence contained in nucleotides 568 to 2045 of SEQ ID NO: 1, or (iii) a complementary strand of (i) or (ii) (page 5, line 1, to page 7, line 7); for comparing the percent identity of the deduced amino acid sequences of the phospholipases to amino acids 20 to 464 of SEQ ID NO: 2 using the Clustal method according to Higgins, 1989, *CABIOS* 5: 151-153 (Example 4); for determining the degree of homology between two nucleic acid sequences using the Wilbur-Lipman method according to Wilbur and Lipman, 1983, *Proceedings of the National Academy of Science USA* 80: 726-730 (page 12, line 29, to page 13, line 8); for producing the phospholipases (Example 5); for purifying the phospholipases and characterizing the properties of the encoded phospholipases (Examples 6-9); for determining phospholipase B activity (page 3, lines 18-24); and how to use the

phospholipase B's encoded by the nucleic acids (page 36, line 11, to page 41, line 30. It is well within the skill of the art to make and use the claimed nucleic acid sequences using Applicant's disclosure.

For the foregoing reasons, Applicant submits that the claims overcome this rejection under 35 U.S.C. § 112. Applicant respectfully requests reconsideration and withdrawal of the rejection.

III. Conclusion

In view of the above, it is respectfully submitted that all claims are in condition for allowance. Early action to that end is respectfully requested. The Examiner is hereby invited to contact the undersigned by telephone if there are any questions concerning this amendment or application.

Respectfully submitted,

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Robert L. Starnes, Ph.D.
Reg. No. 41,324
Novozymes Biotech, Inc.
1445 Drew Avenue
Davis, CA 95616-4880
530-757-8100
530-757-4715 (direct)